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MICROSPOROGENESIS AND -GAMETOGENESIS IN *STRELITZIA REGINAE* AIT.

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Abstract

The anther of *Strelitzia reginae* is tetrasporangiate with a persistent epidermis. The endothecium develops fibrous thickenings, the middle layers are ephemeral and the cells of the glandular tapetum are frequently binucleate.

Divisions of the microspore mother cells are of the successive type and the microspore tetrads are isobilateral, T-shaped and linear.

The spherical, smooth-walled pollen grains are uninucleate at dehiscence.

Uittreksel

MIKROSPOROGENESE EN -GAMETOGENESE IN *STRELITZIA REGINAE* AIT:

Die helmknop van *Strelitzia reginae* is tetrasporangiaat en besit 'n blywende epidermis. Die endotesium ontwikkel veselagtige verdikkings, die middellae gaan verlore en die selle van die klieragtige-tipe tapetum is dikwels tweekernig.

Deling van die mikrospoormoederselle is van die agtereenvolgende tipe en die mikrospoortetrades is isobilateraal, T-vorming en lineêr.

Die sferiese stuifmeelkorrels besit gladde wande en is eenkernig wanneer die helmknop oopspring.

Introduction

Strelitzia reginae Ait. is endemic to South Africa (Phillips, 1951) and is well-known as an ornamental in gardens and parks. The flowers have become very popular in floristry and plants are cultivated for this purpose in many parts of the world. The flower has even been adopted as the civic emblem of Los Angeles.

It is surprising, therefore, that the flowers of this species have rarely been the object of scientific investigation. Apart from the study of Mauritzon (1936) on the embryo-sac development of *S. reginae*, no other work on the embryology of this species could be traced in the literature. Davis (1966) mentions that development of the anther and pollen grains of Strelitziaceae has not been described. In order to gain more information on the embryology of *S. reginae*, a study was made of the microsporogenesis and -gametogenesis of this species.

Material and Methods

Flowers of *S. reginae* were collected from plants growing in Settler's Park, Port Elizabeth. Anthers in different stages of development were fixed and stored in Allen-Bouin type II fluid (Sass, 1958). Dehydration was accomplished in a dioxan-tertiary butyl alcohol series (Sass, 1958) before embedding in wax (Carowax; melting point 54–58°C). Sections were cut on a Jung model 1130 rotary microtome at a thickness of 10 μm. Staining was performed in a Shandon automatic staining machine using the iron-alum-hematoxylin safranin stain of Brooks, Bradley & Anderson (1950). Photographs were taken with a Zeiss photomicrographic camera using Kodak Panatomic X film.

Microspore tetrads were studied by mounting them directly in water, without prior fixation, and viewing under phase contrast. Binucleate microspores were fixed in Carnoy (Sass, 1958), mounted in propionocarmine and viewed under phase contrast.

RESULTS AND DISCUSSION

The anther

In the flowers of *S. reginae* the margins of two of the petals overlap to enclose five stamens and the style (Fig. 1). The anthers are four-lobed and four-loculed (tetrasporangiate). The lobes remain distinct as the anther matures with the result that the two sporangia on each side of the anther do not become confluent before dehiscence.

The epidermis, or outermost layer of the anther, undergoes anticlinal divisions in order to keep pace with the enlargement of the anther. The compact appearance of these cells is maintained up to maturity, at which time the outer walls are covered by a distinct cuticle.

The young wall of the microsporangium consists of a series of concentric layers of cells (Fig. 2). The outermost of these layers, the endothecium, is situated directly beneath the epidermis. Prior to the release of pollen the cells in this layer develop distinctive fibrous bands of thickenings which stretch from the inner tangential walls to the outer walls.

Adjacent to the endothecium are three to six middle layers. These cells all become flattened and disintegrate as the locules increase in size (see Fig. 3) and have become completely degenerated by the time of dehiscence. According to Davis (1966) this phenomenon is due to the fact that the cells of the middle layers generally lack the ability to divide anticlinally with the result that the tissue cannot adjust itself to the multiplying and expanding sporogenous cells within the sporangium.

The innermost wall layer is the tapetum. In most angiosperms, the tapetum consists of a single layer of cells (Maheshwari, 1950) but in *S. reginae* the microsporocytes are commonly separated from the middle layers by two to three layers of tapetal cells (Fig. 2). The probable role of tapetal cells is that of providing nutrients for the developing spores (Foster & Gifford, 1959). The

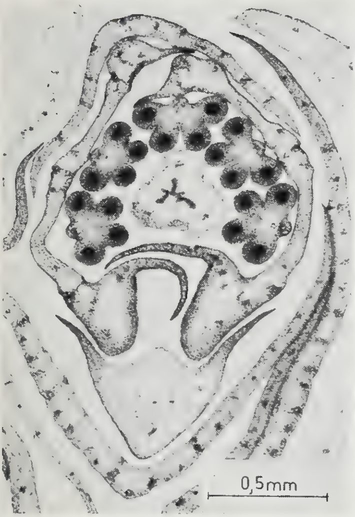

FIG. 1.

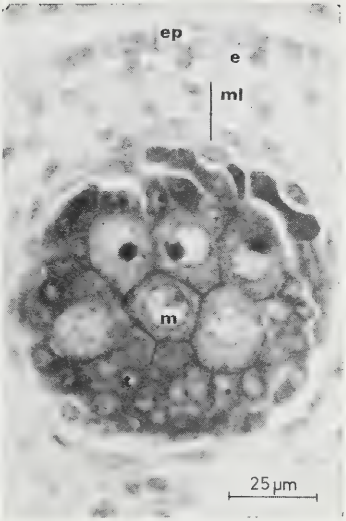

FIG. 2.

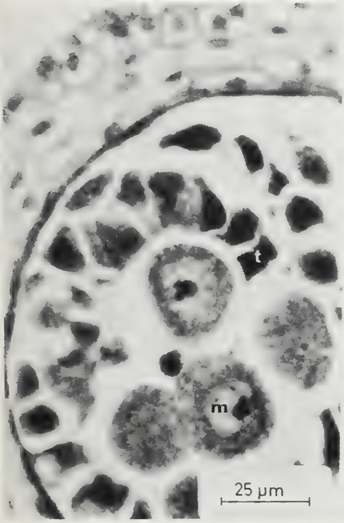

FIG. 3.

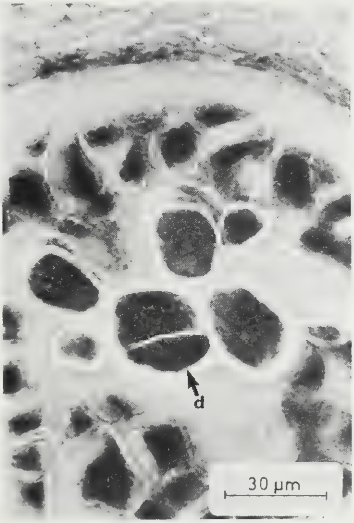

FIG. 4.

FIG. 1.
Transverse section of a young flower of *S. reginae*.

FIGS. 2–4.
Transverse sections of microsporangia of *S. reginae* (d—dyad, e—endothecium; ep—epidermis of anther, m—microsporocyte, ml—middle layers, t—tapetum).

tapetal cells of *S. reginae* are often binucleate. Davis (1966) mentions that such nuclear divisions are a common feature of tapetal cells.

Just prior to the commencement of meiotic divisions in the microsporocytes, the tapetal cells begin to lose contact with the cells of the middle layers and with one another and are finally absorbed. The tapetum is of the glandular or secretory type. This is the type commonly found in angiosperms (Maheshwari, 1950).

Microsporogenesis

The primary sporogenous cells give rise to the microsporocytes which enlarge until they ultimately have a diameter of 25–30 μm (Fig. 3). They are easily distinguished from the surrounding tissue because of their large size and prominent nuclei.

Each functional microsporocyte, by means of meiosis and cytokinesis, gives rise to a tetrad of microspores. The divisions of the microspore mother cells are of the successive type, that is, a cell plate is laid down immediately after the first meiotic division (Fig. 4), and another in each of the two daughter cells after the second division. Although there are frequent exceptions, the successive type is prevalent in monocotyledons (Foster & Gifford 1959).

The arrangement of microspores in the tetrads is determined by the orientation of the homeotypic spindles. According to Davis (1966) the arrangement is commonly tetrahedral, isobilateral or decussate. In anthers of *S. reginae* no tetrads were found having the tetrahedral or decussate arrangements. Tetrads having the isobilateral, T-shaped and linear arrangements were found (Fig. 5) and these were present in approximately equal proportions. It is uncommon to find three types of disposition in the same species and the phenomenon has been encountered in species of only a few genera, including *Musa* (Maheshwari, 1950).

Occasionally abnormal tetrads were encountered. Figure 5d pictures such a "tetrad" containing five cells. Groups of as many as eight cells were found in the anthers examined. This phenomenon is known as polyspory and is generally found only in hybrids characterized by a high degree of sterility (Maheshwari, 1950). Pollen grains arising in this way are reported to be nonfunctional.

Microgametogenesis

The spore members of each tetrad soon separate from each other and lie freely within the microsporangium. Each cell rapidly increases in volume and the accompanying vacuolisation is followed by a displacement of the nucleus from the centre to a place adjacent to the wall.

Division of the microspore gives rise to the vegetative and generative cells (Fig. 6). According to Maheshwari (1950) pollen grains are usually either in the

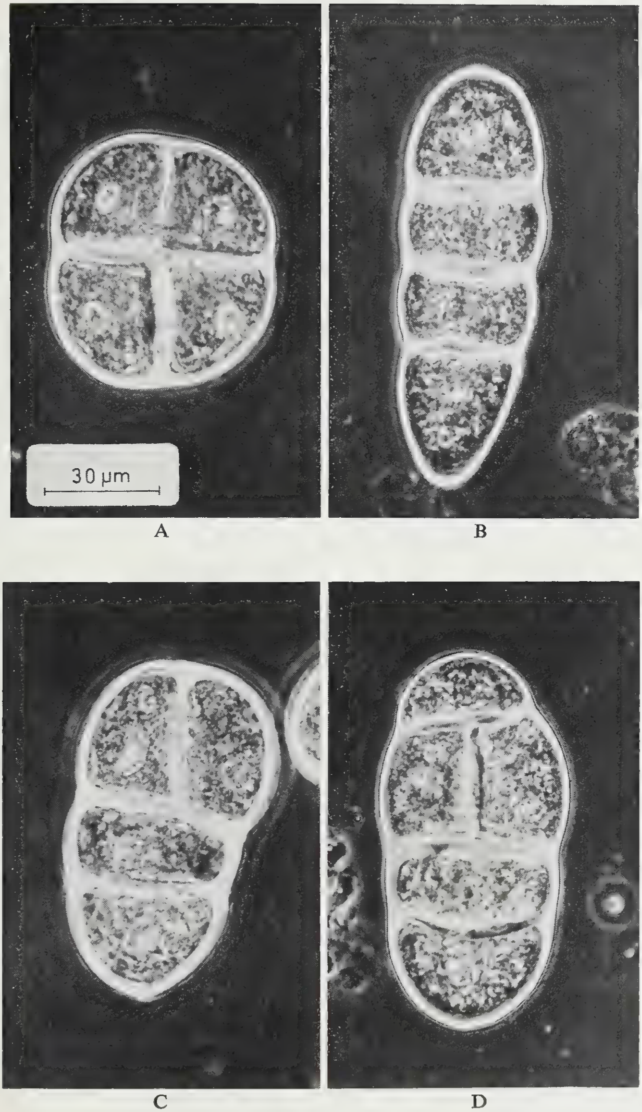

Fig. 5.

Microspore tetrads of *S. reginae*

(*a*) Isobilateral type (*b*) Linear type (*c*) T-shaped type (*e*) Abnormal tetrad containing five spores.

two-celled or three-celled condition when shed, depending on whether the generative cell divides prior to the release of the pollen or not. In the case of *S. reginae*, however, it seems that the vegetative nucleus is short-lived and that it degenerates before the pollen grains reach maturity. All the mature pollen grains examined contained only a single, centrally placed nucleus (Fig. 7). Maheshwari (1950) refers to several species in which the vegetative nucleus is known to degenerate before the pollen grains start to germinate and cites this as evidence against a possible role of this nucleus in directing growth of the pollen tube.

Attempts were made to investigate division of the generative nucleus after germination of the pollen grain. Pollen grains of *S. reginae* can be successfully germinated on an agar medium containing 10 to 15 per cent sucrose, 0,008 per cent boric acid and an aqueous extract of stigma tissue. Various nuclear stains were tested and in all cases the cytoplasm stained intensely and obscured the nuclear material.

The mature pollen grain (Fig. 7) has a dense cytoplasm. It is spherical in shape and has a thick exine and thin intine. The exine is unsculptured and no furrows or pores were observed in this layer. Erdtman (1972) has also observed that the pollen grain of *S. reginae* is nonaperturate.

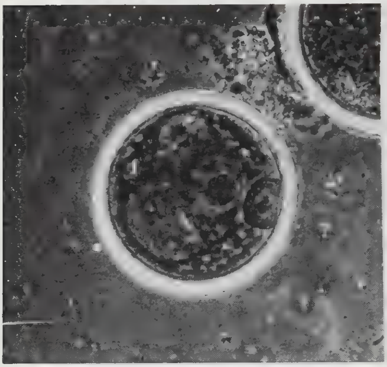

FIG. 6.
Microspore of *S. reginae* in the two-celled stage.

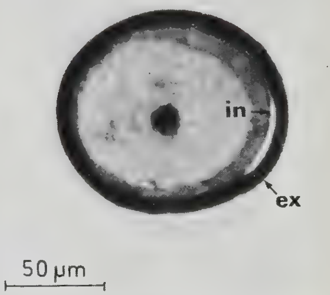

FIG. 7.
Uninucleate pollen grain of *S. reginae* (ex—exine, in—intine).

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